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File: USPT

Jun 1, 1993

DOCUMENT-IDENTIFIER: US 5215882 A

TITLE: Method of immobilizing nucleic acid on a solid surface for use in nucleic acid hybridization assays

Abstract Text (1):

Methods of immobilizing nucleic acid on a solid surface for use in nucleic acid hybridization assays is disclosed. The methods of the invention comprise reacting a modified nucleic acid strand comprising a variable portion and an anchor portion wherein the variable portion comprises a nucleotide sequence having a selected base sequence and the anchor portion comprises at least one nucleotide base modified with a primary amine function or nucleotide base equivalent having a primary amine function and reacting the modified nucleic acid strand with a free aldehyde group of the solid surface in the presence of a reducing agent to form complexes of the modified nucleic acid strand and at least a portion of the free aldehyde groups on the solid surface.

Brief Summary Text (10):

The methods of the invention provide methods of immobilizing nucleic acid to a solid surface having a free aldehyde group for use in nucleic acid hybridization assays. In the methods of the invention a modified nucleic acid strand comprising a variable portion and an anchor portion wherein the variable portion comprises a nucleotide sequence having a selected base sequence and the anchor portion comprises at least one nucleotide base modified with a primary amine function or nucleotide base equivalent having a primary amine function is reacted with the free aldehyde group of the solid surface in the presence of a reducing agent to form complexes of the modified nucleic acid strand and at least a portion of the free aldehyde groups.

Detailed Description Text (13):

To reduce background interference in a hybridization assay, aldehyde groups on the solid surface that did not react with the modified nucleic acid strand may be optionally modified by reaction with an amino acid, such as .alpha.-amino caproic acid in the presence of a reducing agent. This step will convert the remaining aldehyde groups to acidic functions by reaction with the acid, so that the aldehyde groups will not be available to bind with nucleic acid or other reagents in nucleic acid hybridization assays and also introduces a negative charge on the surface, which aids in keeping background interference low. Conversion of unreacted aldehyde groups may readily be accomplished after immobilization of the modified nucleic acid strand by applying a mixture of 0.1 M aminocaproic acid and 0.1 M sodium cyanoborohydride to the solid surface. The mixture may be applied to the solid surface by soaking the solid surface in a solution containing .alpha.-amino caproic acid and sodium cyanoborohydride, spraying or any other suitable method. After approximately one-half to two hours the paper is washed with water few times to remove all reagents and finally with phosphate buffered saline (PBS) and dried and stored.